ESTABLISHING THE DESIGN SPACE OF A FILTRATION-BASED OPERATION FOR THE CONCENTRATION OF HUMAN PLURIPOTENT STEM CELLS

Manuel JT Carrondo, iBET/FCT-NOVA
Apartado 12, Oeiras, Portugal
mjtc@ibet.pt
Bárbara Cunha, iBET/ITQB-NOVA
Ricardo J.S. Silva, iBET/ITQB-NOVA
Cláudia Correia, iBET/ITQB-NOVA
Alexey Koshkin, iBET/ITQB-NOVA
Paula M. Alves, iBET/ITQB-NOVA
Margarida Serra, iBET/ITQB-NOVA
Cristina Peixoto, iBET/ITQB-NOVA

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Stem cell manufacturing is often very challenging due to the complexity of the biological system. Quality-by-design (QbD), a risk-based framework based on relating process parameters and product quality, can cope with such complexity in process design [1] aiding to develop robust and reproducible unit operations.

This work describes a shortcut approach for the design of tangential flow filtration for the concentration of human induced pluripotent stem cells (hiPSC), supported by design of experiments (DoE) approach.
First, critical quality attributes (CQA), corresponding to the characteristics that ensure the final product quality, and critical process parameters (CPP), which directly affect cells' CQA, are identified. Thereafter, a design space is developed, studying how a range of variability in CPP allows to achieve CQA [2]. Thus, CPP of shear rate, permeate flux and cell load were considered, and initially their impact on hiPSC recovery yield and viability responses was studied.

A full factorial design confirmed significant interaction effects between all CPP, affecting both responses. The developed statistical model predicted that high shear rate (3000 s^{-1}), permeate flux (250 LMH) and medium cell load (2 x 10^6 cell/cm^2) would maximize both cell recovery yield and viability, where over 80% of hiPSC would be recovered after a volume reduction factor of 20 with high viability (over 93%). Such conditions were validated experimentally, and by performing a robustness analysis, the success rate of these operating conditions was assessed (65 - 70%). A parametric study was then conducted, identifying that increasing the shear rate (up to 3370 s^{-1}) allowed to achieve the specified requirements for cell recovery yield (> 80%) and viability (> 90%) in 100% of the cases and no impact in hiPSC's CQA in terms of identity, proliferation capacity and pluripotency was observed.

References
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